REMARKS/ARGUMENTS

Claims 1-31 are pending. Claims 1, 4-6, and 9 are presently under examination and stand rejected. In the instant amendment, claims 1 and 4 are amended. Reconsideration of the claims is respectfully requested.

According to the Advisory Action mailed August 12, 2003, the Applicant has not yet sufficiently addressed the combination of the Rabinovitch and Lenschow references, or Rabinovitch's discussion of specific immune stimulation. In response, Applicant now provides additional evidence and explanation.

Rejection Under 35 U.S.C. §112, second paragraph

Claim 4 was rejected as allegedly indefinite on the grounds that "the administered substance" has no antecedent in claim 1, from which claim 4 depends. Claim 1 has been amended to refer to administering to the subject an effective amount of an anti-CD28 agonist selected from a defined group of agonists. Support for administration of an anti-CD28 agonist can be found in the application as filed, for example in original claim 1.

Claim 4 has been amended to be consistent with the amended language of claim 1.

It is respectfully submitted that these amendments overcome the Examiner's objection.

Rejection Under 35 U.S.C. §103(a)

Claims 1, 4-6, and 9 were rejected as allegedly obvious over Rabinovitch, Diabetes, 43:613-621 (1994) [Rabinovitch] and Lenschow et al., Immunity, 5:285-293 (1996) [Lenschow '96; IDS-Y], in view of either King et al., Eur. J. Immunol., 25:587-595 (1995) [King; IDS-W] or Webb et al., Blood, 86:3479-3486 (1995) [Webb; IDS-AQ].

In general, amended claim 1 is drawn to a method of preventing the development of diabetes by administering a T cell co-stimulatory receptor CD28 agonist. Lenschow '96 reports that inhibition of CD28 signaling leads to increased diabetes and decreased Th2 response. Rabinovitch discusses modulation of the immune system via an increased Th2 response to treat diabetes. King and Webb discuss a CD28 agonist that leads to increased Th2 response.

As noted in MPEP 2142, a *prima facie* case of obviousness requires at least (1) some suggestion or motivation, either in the cited references themselves or in the knowledge generally available to the artisan, to modify the reference or to combine reference teachings, and (2) a reasonable expectation of success. What is more, MPEP 2143.01 requires that where references provide conflicting information, the Office must weigh the power of each reference to suggest solutions to the artisan. It is respectfully submitted that the cited references fail to meet these requirements.

I. The Combination of Rabinovitch and Lenschow '96 is Improper

Rabinovitch and Lenschow '96 are each cited for the same proposition: that Th2 stimulation can be used to treat diabetes. Yet a full reading of each of these references, particularly in light of the state of the technological field, reveals that each is deficient in supporting this proposition, and further that the combination of the references is similarly deficient and improper.

A. Lenschow '96 report of CD28 signal timing is unclear

The Office has suggested that conflicting results in the literature regarding the CD28 pathway signaling and immunosuppression can be explained by the timing of the CD28 signal. Applicants respectfully disagree, and draw to the Office's attention the following references which indicate the understanding of the artisan shortly prior to and at the priority date of the subject invention (October 1, 1997). Copies of these references, where not previously provided, are enclosed.

MPEP 2143.01 requires that all teachings in the literature must be considered and where the teachings of two or more references conflict, the Examiner must weigh the power of each reference to suggest solutions to one of ordinary skill in the art, considering the degree to which one reference might actually discredit another. The one consistent theme in the cited references is the reported use of CD28 antagonists in preventing the development of autoimmune diseases, including IDDM.

Lenschow et al., in Science, 257:789-792 (1992) [Lenschow '92] report administration of CTLA-4Ig, as an antagonist to block CD28 signaling, can have

immunosuppressive effects in treating graft rejection. Finck et al., in Science, 265:1225-1227 (1994) [Finck] also report an immunosuppressive effect of CD28 blockade with CTLA-4Ig to treat the autoimmune disease lupus. The importance of a CD28 signaling timing is discussed in Lenschow et al., in J. Exp. Med., 181:1145-1155 (1995) [Lenschow '95], where it is reported that treatment of NOD mice at two to four weeks of age, either with CTLA-4Ig or with anti-B7-2 monoclonal antibody (CD28 antagonists), prevent the development of IDDM, and treatment at a later age has no effect. Here, Lenschow '95 concludes, at page 1153, col. 2:

> "these results clearly demonstrate that *T cell co-stimulation* is an *essential* component of the in vivo activation of autoreactive T cells and the *development of autoimmune diabetes*". (Emphasis added).

These studies suggest that activation of CD28 signaling results in the development of IDDM, and that antagonists of CD28 signaling may be useful in treating autoimmune diseases. Yet in contrast to the Lenschow '95 report that the timing of the CD28 signaling blockade is most effective at two to four weeks, Lenschow '96 reports an opposite result: that a signaling blockade at two to five weeks is ineffective.

Lenschow '96 discusses two methodologies of studying CD28 signaling. In a first methodology, at page 285, right col., referring to unpublished data, CTLA-4Ig treatment to block CD28 signaling is reported to be only efficacious in preventing diabetes when administered to mice between 5 and 7 weeks of age. In a second methodology, at page 290, right col., in a study on CD28-/-mice, the critical window for disrupting CD28 signaling is hypothesized to be in the first two weeks, in order to see an increased rate of disease onset.

Thus, at or about the time the present application was filed, reports by those of skill in the art could be summarized as follows:

1. CD28 antagonists are reported as immunosuppressive (Lenschow '92; Finck).
2. CD28 antagonists are effective only at 2-4 weeks (Lenschow '95).
3. CD28 antagonists are effective only at 5-7 weeks (Lenschow '96).
4. CD28 knockout mice developed disease (Lenschow '96).

These results present at least two contradictions. First, the inhibition of CD28 signaling by one method, administration of the antagonist CTLA-4Ig, was reported to prevent

diabetes, but it is unclear from the literature whether this is effective at two to four weeks, or at five to seven weeks. Second, in contradiction to the antagonist treatment approach, it is reported that by disruption of the pathway in CD28-/-mice, diabetes was actually promoted. Thus, there are conflicting results in the references, and these cannot be explained by the timing of the CD28 signal. What is more, these reports also conflict with other subsequent reports discussing the prevention of diabetes with CD28 antagonists.

For example, Levisetti et al., J. Immunol., 159:5187-5191 (1997) writing shortly after the priority date of this application and therefore reflecting what was still the view of those of skill in the art even after the date of this invention, describe the use of the CD28 antagonist, CTLA-4Ig, to suppress the immune response of non-human primates to allogeneic pancreatic islet transplantation. They conclude that CD28 co-stimulation is important for development of the immune response. And later, Greenfield et al., Crit. Rev. Immunol., 18(5):389-418 (1998), in reviewing the current understanding at that time for CD28 costimulation, report that blocking the CD28 pathway can result in immunosuppression with implications for treating autoimmune diseases, whereas activating the CD28 pathway may be useful for stimulating the immune system for tumor elimination. Neither of these two subsequent publications even remotely suggests a therapeutic role for CD28 agonists.

The picture then for the artisan, regarding CD28 signal impairment, at the date of the invention is as follows: (1) studies using CD28 antagonists consistently showed suppression of immune responses both in the context of graft rejection and in the context of autoimmune diseases, including autoimmune diabetes, although the effective agonist treatment window was reported as 2-4 weeks in one study and 5-7 weeks in another study; (2) studies with CD28 knockout mice showed an increased rate of disease onset, and the hypothesized critical window was the first two weeks of life. In sum, when considered as a whole, the literature presents a confusing picture regarding the timing and effectiveness of antagonist therapy.

B. Inference that Lenschow suggests agonist therapy is improper

The Office acknowledges that Lenschow '96 fails to teach agonist therapy, but suggests that because inhibition of CD28 signaling during the first two weeks of life is reported

to exacerbate IDDM, the artisan could infer an implicit disclosure that the opposite strategy of stimulating CD28 signaling during this time period would induce a Th2 response and protect from the development of diabetes. Yet no support is provided by the Office for this suggestion.

Applicant has, however, submitted a Declaration of one skilled in the art that Lenschow does not suggest agonist therapy. As noted in the Declaration at ¶¶9-12, the artisan would conclude that there is a likelihood that other co-stimulator molecules were compensating for the lack of CD28 in the knockout animals, as noted by Lenschow themselves and in view of the fact that the critical autoantigen-sensitive T cells were able to develop in the absence of CD28 signaling.

As noted in MPEP 2141.01 III, the content of the cited references must be determined at the time the invention was made, in order to avoid impermissible hindsight. The Examiner must caution against selecting only those references which support his or her position, while ignoring other references which negate that position. The Examiner's position, namely that once one has shown that elimination of CD28 signaling from birth accelerates diabetes development, one of skill in the art would automatically then assume that stimulation of CD28 signaling would prevent diabetes development, is an improper application of hindsight and fails to eliminate from consideration the proven results of the presently claimed invention. If the Examiner limits her consideration to the confused state of the art at the date of the invention, such a conclusion is not automatic.

It is entirely speculative to conclude that, because the literature reports an absence of CD28 signaling leads to an exacerbation of autoimmune diabetes, treatment with an agonist of the pathway would therefore be therapeutic, particularly since it has already been reported in the literature that antagonists are efficacious in preventing IDDM. In view of the above-noted complexities and contradictions in the field, it is respectfully submitted that no one of skill in the art could possibly have concluded that stimulating CD28 signaling would have an effect opposite to the effect noted by Lenschow '96. If the Examiner's proposition had occurred to one of ordinary skill in the art, it would have been no more than mere speculation that such an approach might be "worth a try". Even if it were accepted that one of skill in the art would be of this view,

which is not conceded by the Applicant, this would not rise to the level required to demonstrate obviousness.

According to the Office Action, despite the fact that Lenschow '96 discusses only CD28 antagonists, this does not mean that the artisan at the time the invention was made would not have readily appreciated that during the pre-diabetic stage, when blocking CD28 signaling is reported to exacerbate disease, an antibody that agonizes CD28 signaling would be desirable. The Examiner has presented no evidence that one of ordinary skill in the art would have that appreciation and, in fact, the applicant has pointed to other pieces of art which would predispose one of ordinary skill in the art at that time not to expect success by stimulating CD28 signaling. As noted in the aforementioned MPEP 2141.01 III,

> "it is difficult but necessary that the decision maker forget what he or she has been taught ...about the claimed invention and cast the mind back to the time the invention was made (often as here many years), to occupy the mind of one skilled in the art who is presented only with the references and who is normally guided by the then-accepted wisdom in the art".

Lenschow's own studies had shown success in preventing IDDM by using CD28 antagonists, and hence an individual of ordinary skill within the art would not have anticipated that an agonist would yield a similar efficacy in the prevention of IDDM.

In sum, there is simply no suggestion for an immunosuppressive therapeutic role for a CD28 agonist in the references of record. Among all of the cited references, not once is a potential therapeutic role for CD28 agonists mentioned in the context of treating IDDM or any other autoimmune disorder. The artisan would not have concluded, and in fact there is no evidence that one of skill in the art did conclude, that the results shown by Lenschow '96 could be reversed by an additional co-stimulatory function such as CD28 activation.

C. Rabinovitch discusses specific tolerance but fails to teach T cell activation

Rabinovitch is cited by the Office for the proposition that Th2 stimulation can be used to treat diabetes. Yet a close reading of this reference reveals this conclusion to be incomplete with respect to the presently claimed invention.

Rabinovitch fails to provide any teaching regarding the CD28 signaling pathway. Instead, Rabinovitch et al. tries to reconcile the inconsistent findings that both immunosuppressive and immunostimulatory approaches can treat diabetes. Firstly, Rabinovitch promotes the use of microbial adjuvants as therapy for IDDM onset, which has been clearly refuted in the literature using clinical studies as has been elucidated fully in prior submissions by the Applicant. Secondly, Rabinovitch suggests specific immune stimulation by administration of GAD65, an islet beta-cell autoantigen itself considered to be involved in the development of autoimmune diabetes. Rabinovitch notes that prevention of autoimmune diabetes in NOD mice by administration of GAD65 was associated with the induction of specific tolerance to this polypeptide. This was therefore not immune stimulation or T cell activation but rather induction of anergy, or T cell hyporesponsiveness, by administration of a specific polypeptide that is a known antigen in the onset and progression of autoimmune diabetes.

That Th1 cell hyporesponsiveness, or inactivation, is associated with the prevention of IDDM is also reported in Kaufmann et al., Nature, 366:69-72 (1993) (copy enclosed) at page 72, "...the absence of T-cell reactivity to GAD..." is responsible for the mitigation of autoimmune diabetes in this context, rather than an upregulation of Th2 immune function or its associated cytokines. In light of Kaufmann's findings, one of skill in the art would have interpreted that Rabinovitch's GAD-mediated effects as likely due to T cell anergy of Th1 cytotoxic functions rather than the induction of a regulatory Th2 function.

Induction of anergy by administration of a specific autoantigen to which an animal has become sensitized does not involve CD28 signaling and thus does not suggest to one of ordinary skill in the art that autoimmune diabetes could be prevented by stimulation of CD28 signaling. Furthermore, Rabinovitch does not indicate that GAD-specific anergy mediated by this approach requires the induction of Th2 function. Rabinovitch's comments on immunostimulation encouraging a Th2 response would therefore not be linked by one of skill in the art to the use of an autoantigen to produce anergy.

According to the Office, Rabinovitch describes immuno-stimulatory procedures to prevent IDDM (auto immune diabetes) in the NOD mouse via T cell differentiation along a

protective Th2 pathway and uses this as a basis for considering immunostimulation in humans to prevent diabetes. Yet, as noted above, Rabinovitch does not describe immunostimulatory signals leading to T cell activation, but rather tolerogenic signals leading to T cell hyporesponsiveness, which does not involve the upregulation of CD28 signaling as described *supra*. Thus, the specific immune stimulation by administration of GAD65 reported by Rabinovitch does not involve CD28 signaling nor does it require Th2 induction.

D. No motivation to combine Rabinovitch and Lenschow '96

As noted above, Lenschow '96, when viewed in context with other literature references, is confusing regarding the timing and effectiveness of antagonist therapy, and provides an inconclusive report with CD28 knockout mice. No role for CD28 agonist therapy is taught or suggested. What is more, Rabinovitch reports T cell hyporesponsiveness which does not involve CD28 signaling.

Thus, Lenschow '96 paints a confusing and incomplete picture of CD28 signaling, and Rabinovitch fails to even suggest a therapeutic role for the CD28 pathway. It follows that there would then be no motivation to combine these references. Rabinovitch's teachings regarding treatment of diabetes with non-specific immune stimulation has been largely discredited and his discussion of administering a relevant auto-antigen does not suggest stimulation of the CD28 pathway. Assuming, *arguendo*, that one of skill in the art could have found an inference in Rabinovitch for the supposition of stimulating a Th2 response and reducing a Th1 response, he or she would not have been motivated to combine that possibility in Rabinovitch with a complete 180 degree reversal of the approach reported to be successful by Lenschow '96 in combating IDDM development, namely inhibition of CD28 signaling by using CD28 antagonists.

What is more, to the extent that these references discuss Th2 cell response, from Lenschow '96 there is no suggestion that CD28 agonist would effect Th2 induction, and from Rabinovitch the report is that tolerogenic signals lead to T cell hyporesponsiveness, which is entirely different from immunogenic signals leading to Th2 induction. Put simply, the references themselves do not suggest the combination, as is required by the MPEP.

There is no motivation for one of skill in the art to combine the teachings of Rabinovitch with Lenschow '96 rather than with any of the other references discussed above, which emphasized inhibition of CD28 signaling.

The Examiner has reviewed the Declaration of Dr. Delovitch but has held that it is insufficient overcome this rejection. In the Advisory Action, the Examiner comments that the Declaration "does not address what the ordinary artisan at the time the invention was made would have found obvious in view of the combination of the references". It is submitted that there is no onus on the applicant to describe what one of skill in the art would have found obvious.

E. Lenschow '96 and Rabinovitch fail to provide reasonable expectation of success

According to MPEP 2143.02, in order to establish a reasonable expectation of success, at least some degree of predictability is required, and predictability is determined at the time the invention was made.

Those authors were people of ordinary skill in the art, as is Dr. Delovitch who has also provided evidence that in his view, one of skill in the art would not have had a reasonable expectation of success, at the date of the invention, in treating or preventing IDDM by up regulating the Th2 arm of the immune response or by attempting to use stimulation of CD28 signaling.

In a novel departure from the literature, the Applicants now present evidence that CD28 agonist treatment completely prevented the development of IDDM (see application at page 27, lines 12-14). Thus, this approach uses the same animal model as described in Lenschow '95, but precisely the opposite strategy: treatment with an agonist instead of an antagonist.

II. The Combination of Rabinovitch and Lenschow '96 with King or Webb is Improper

According to the Office, King and Webb show that the ordinary artisan at the time the invention was made would have recognized that an antibody could be used to stimulate CD28 and that this stimulation would result in a Th2 type of response.

The Applicant respectfully disagrees. Both of these studies report the effect of anti-CD28 antibodies on unprimed, naïve T cells in vitro. Firstly, as well known to one of skill in the art, a number of agents have been demonstrated to generate Th1 or Th2 cells *in vitro*, but have not proved to be efficacious in this regard *in vivo*. An example of such a discrepancy is illustrated by studies on IL-12 receptor blockade (Piccotti et al., (1997), J. Immunol. v. 158, pp. 643-648).

Secondly, King utilized peripheral blood mononuclear cells from normal healthy individuals, while Webb utilized naïve human CD4+ cells from umbilical cord samples. The artisan would not have concluded that a material which could lead to increased Th2 function in naïve T cells would produce a reversal of phenotype in already activated Th1-type cells, such as the cells which are involved in the development of autoimmune diabetes. The artisan would not have anticipated such a reversal *in vitro*, far less in the much more complex situation occurring *in vivo*. It is therefore submitted that one of skill in the art would not have considered King or Webb as relevant to the *in vivo* control of autoimmune diabetes. He or she would therefore have had no motivation to apply the antibodies of King or Webb to the problem of diabetes prevention, nor would there have been any expectation of success based on these references.

It is respectfully submitted that, for all of the above reasons, the presently pending claims of this application are not obvious in view of the cited references.

CONCLUSION

In view of the foregoing, Applicants believe all claims now pending in this Application are in condition for allowance and an action to that end is respectfully requested.

If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 650-326-2400.

Respectfully submitted,

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Attachments

Lenschow et al., (1992), Science, v. 257, pp. 789-792
Finck et al., (1994), Science, v. 265, pp. 1225-1227
Lenschow et al., (1995), J. Exp. Med., v. 181, pp. 1145-1155
Levisetti et al., (1997), J. Immunol., v. 159, pp. 5187-5191
Picotti et al., (1997), J. Immunol., v. 158, pp. 643-648
Kaufmann et al., (1993), Nature, v. 366, pp. 69-72

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